Tumor-derived antibodies for multifaceted immunotherapeutic targeting of human ovarian cancer

DISCLOSURES

Consultant for: Compass Therapeutics (until 2020), Anixa, Leidos, Alloy Therapeutics, Radyus Research.

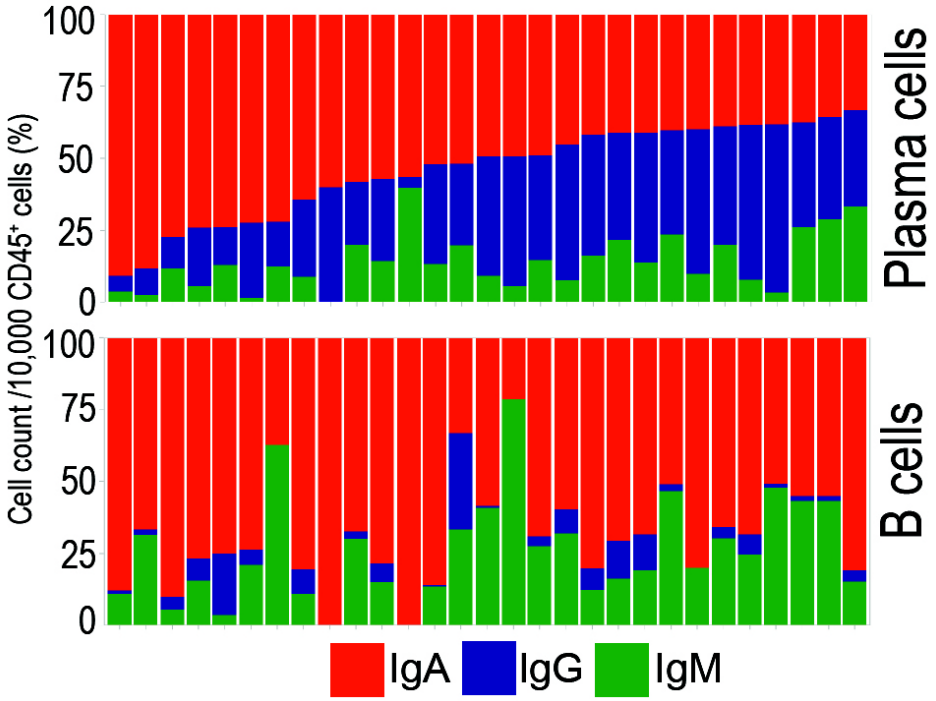
Grant/Research support from: Compass Therapeutics (until 2020), ITUS.

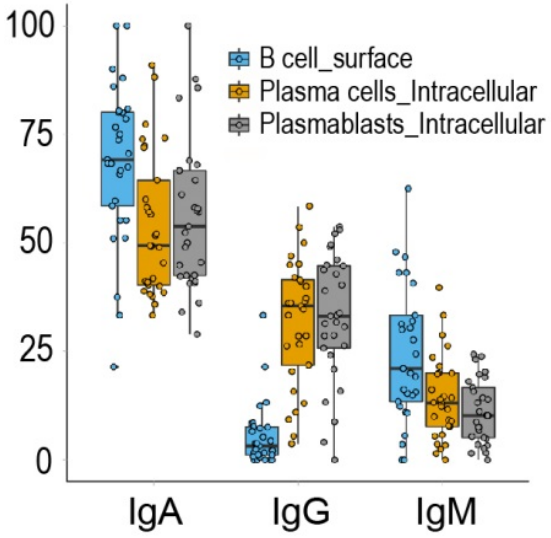
Stock options in: Compass Therapeutics, Anixa Bioscience, Alloy Therapeutics.

Honoraria from: Compass Therapeutics (until 2020), Anixa, Leidos, Alloy Therapeutics

Jose R Conejo-Garcia, MD, PhD, Ab Society/OCRA, September 2021

IgA and IgG dominate Ab secretion by tumor-associated plasmablasts in most ovarian carcinomas





Plasma cell: CD45+CD3-CD20-CD38+CD138+
Plasmablast: CD45+CD3-CD19+CD20-CD38highCD27high
B cell: CD45+CD3-CD19+CD20+

Biswas et al; Nature; 2021

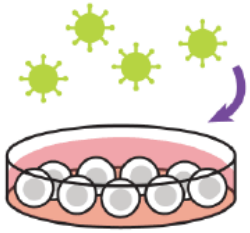
What are the specificities of antibodies produced by TLS^{+} tumor-associated B cells?

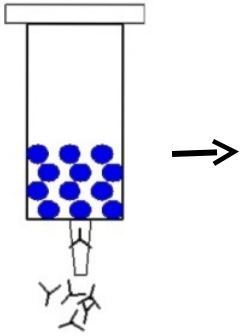
Cloning ovarian cancer-reactive B cells

Established bank of viable dissociated tumor cells

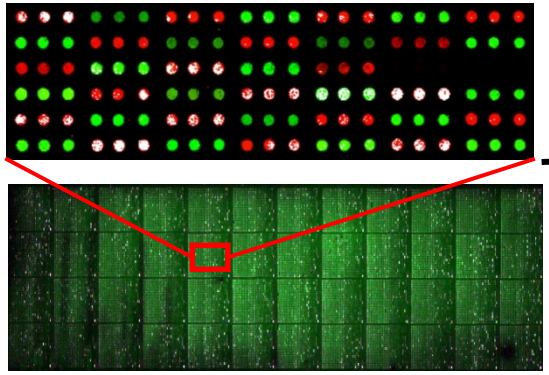
Separation of viable B cells from TLS+ tumors

CD40/IL-21 activation + EBV immortalization

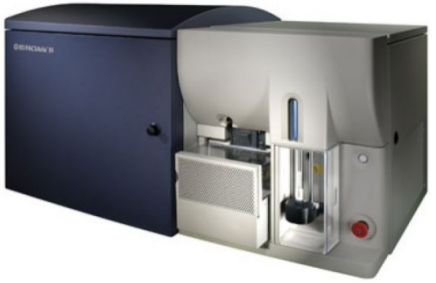




Antibody purification from supernatants

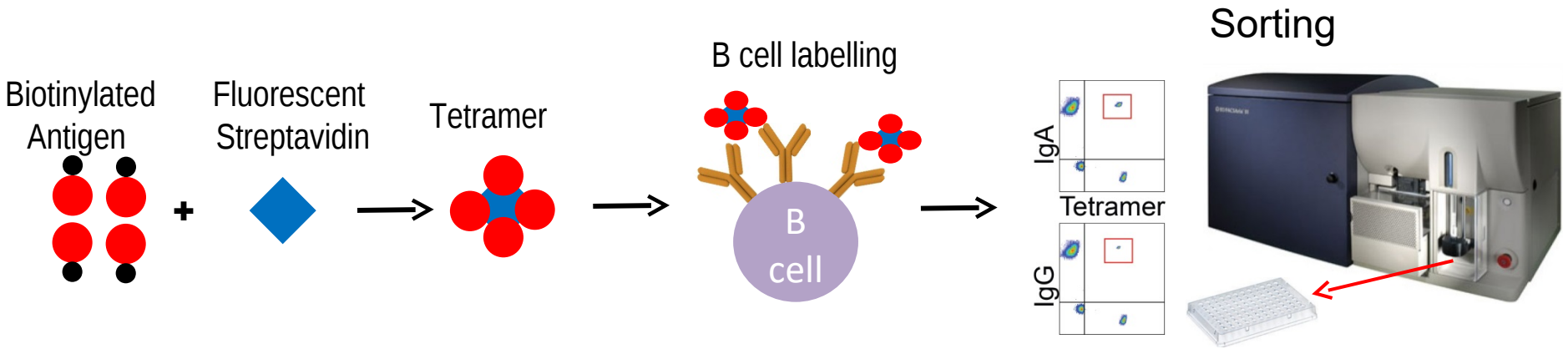


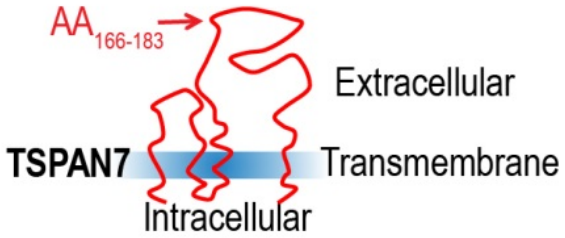
Screening of reactivities using Proteome arrays

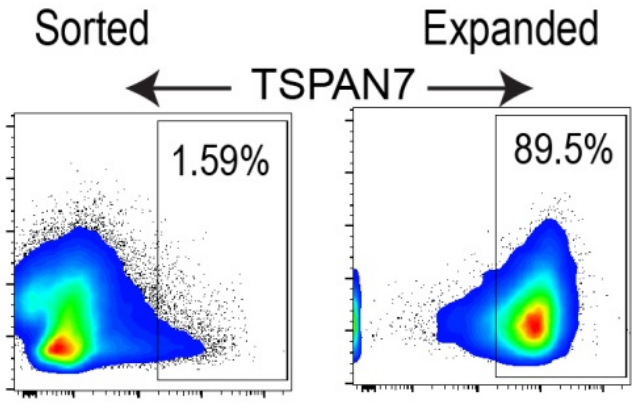


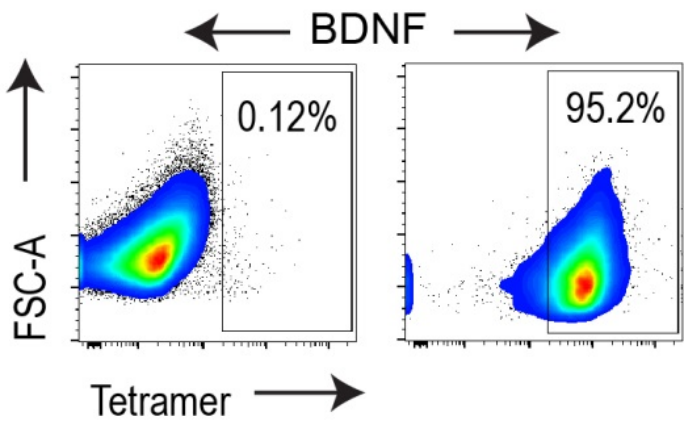
Sorting of reactive B cells using tetramerized biotinylated peptides

Sorting of B cells reacting against extracellular domains using biotinylated peptides



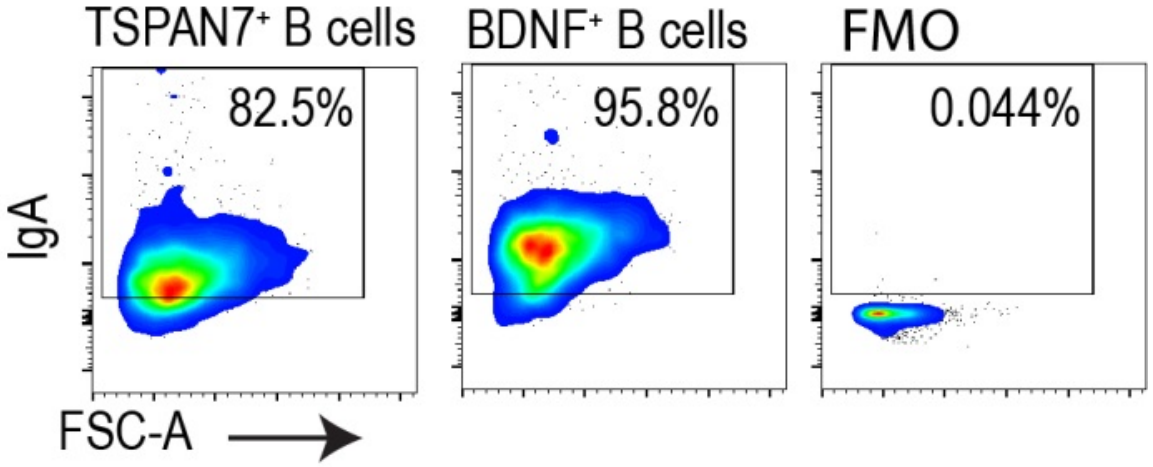


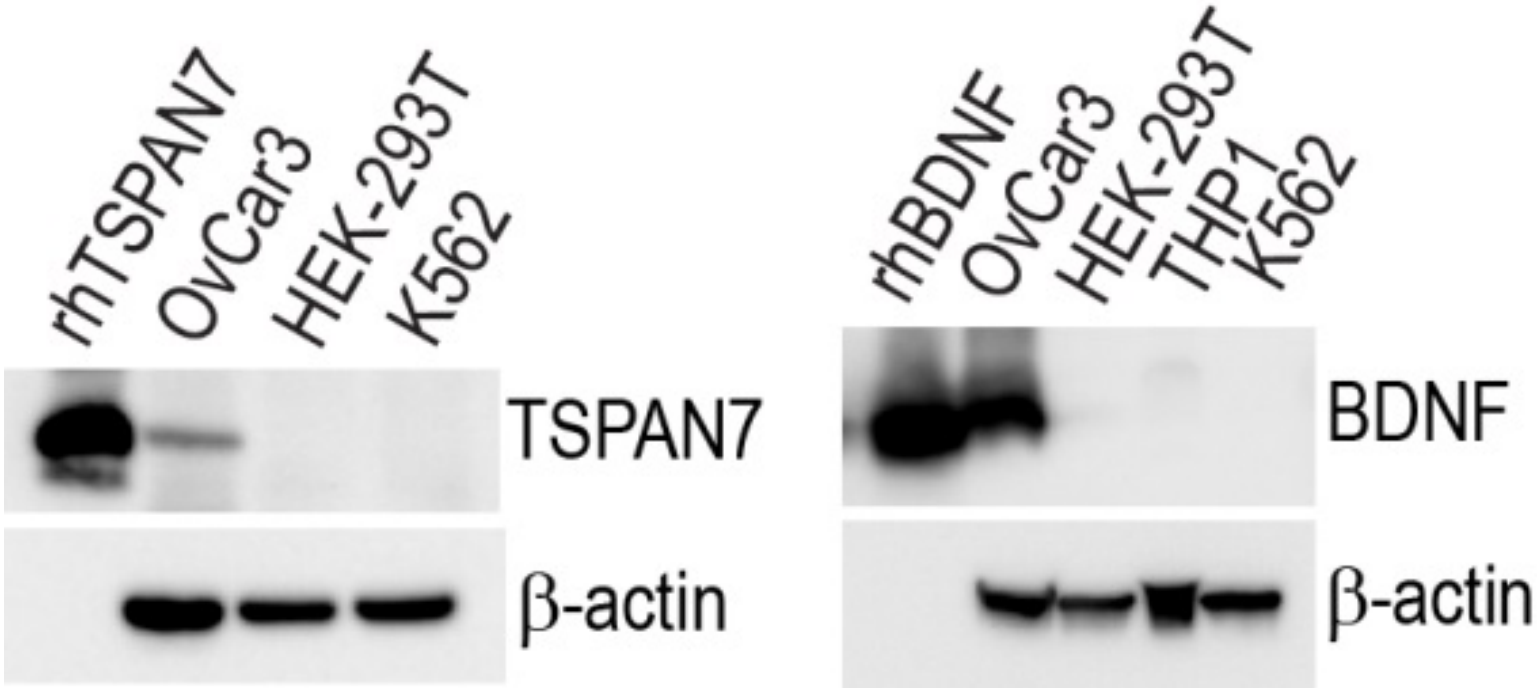




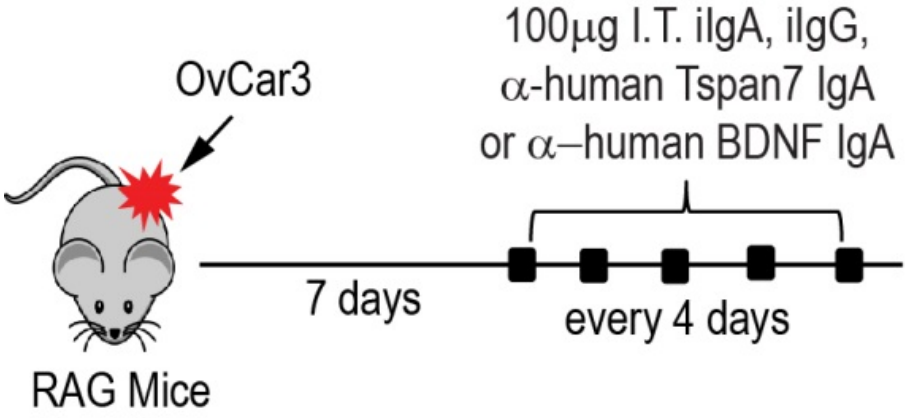
Biswas et al; Nature; 2021

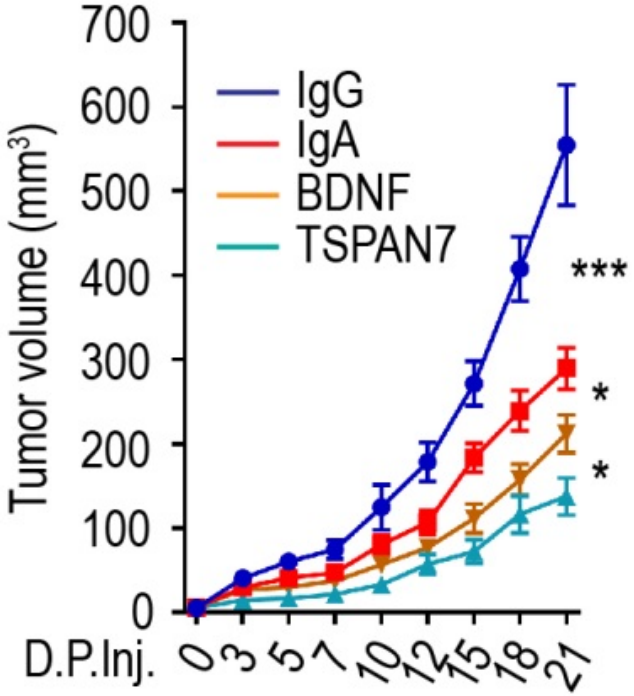
Expanded B cells reacting against extracellular domains using biotinylated peptides retain specificity

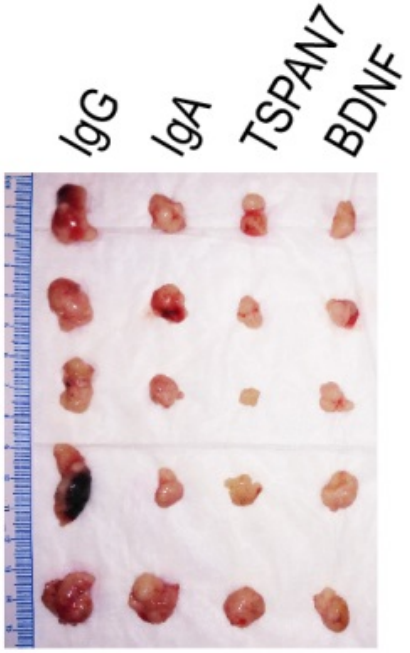


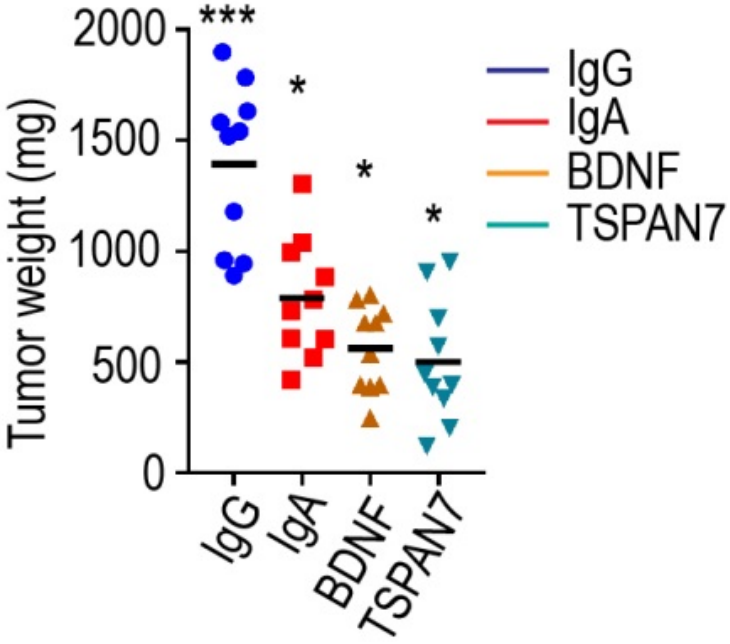


Tumor-derived IgA antibodies exert immune pressure against tumor growth

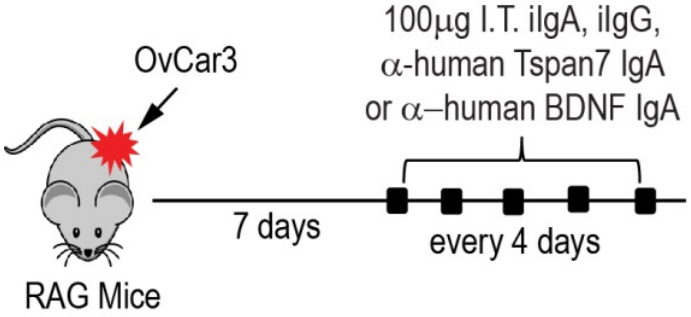


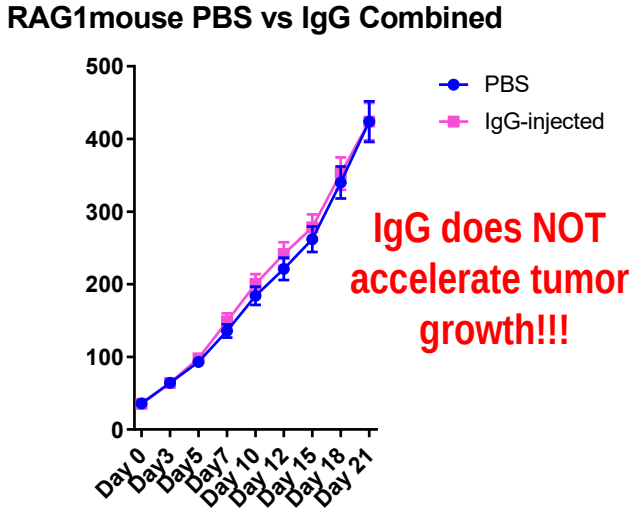




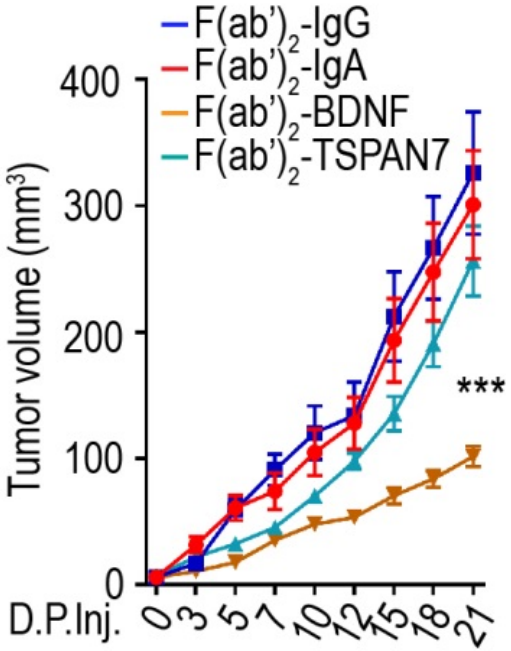


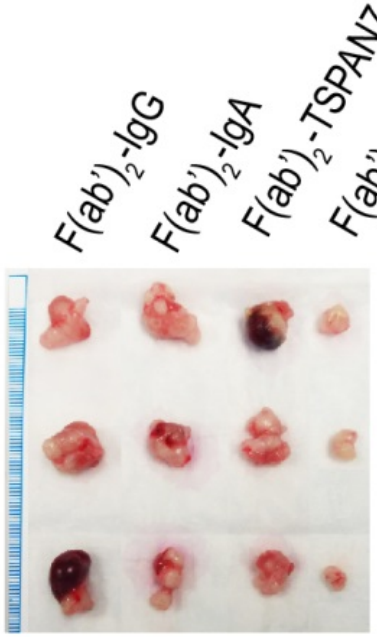
Tumor-derived IgA antibodies exert immune pressure against tumor growth through different mechanisms

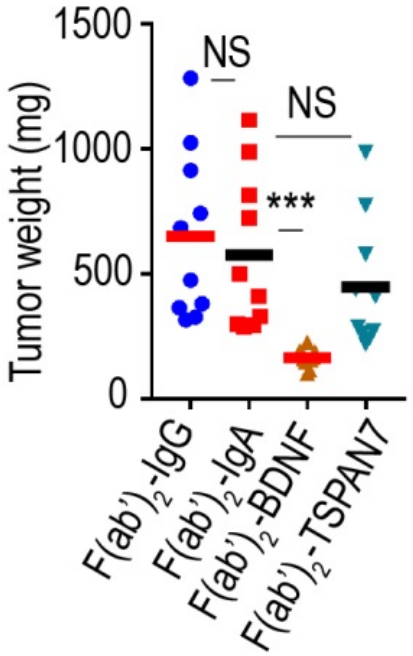




IgG does NOT accelerate tumor growth!!!

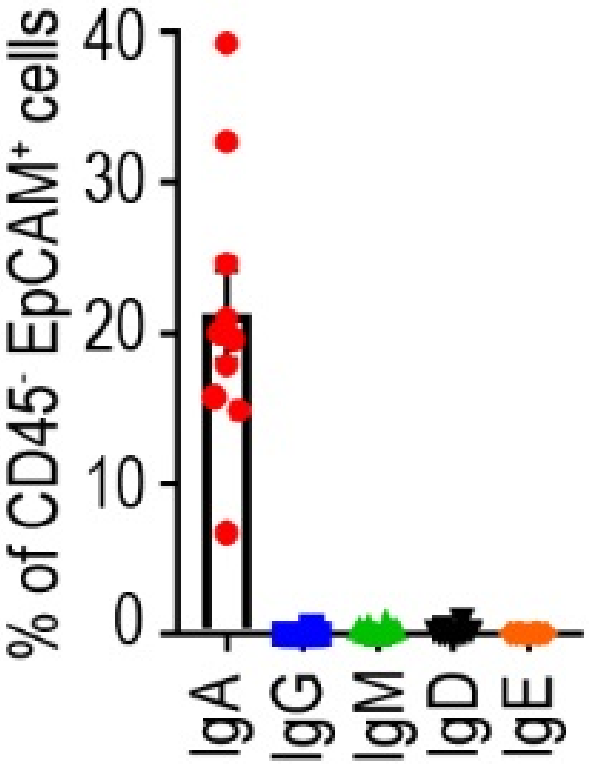


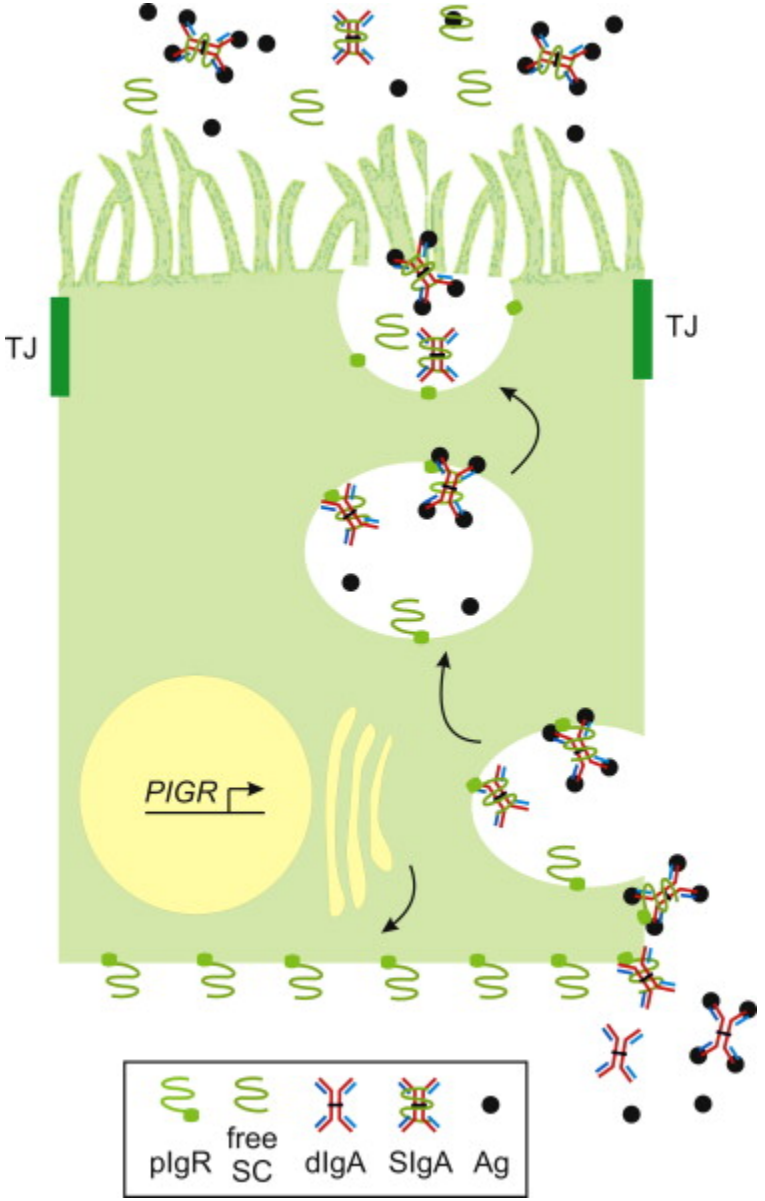




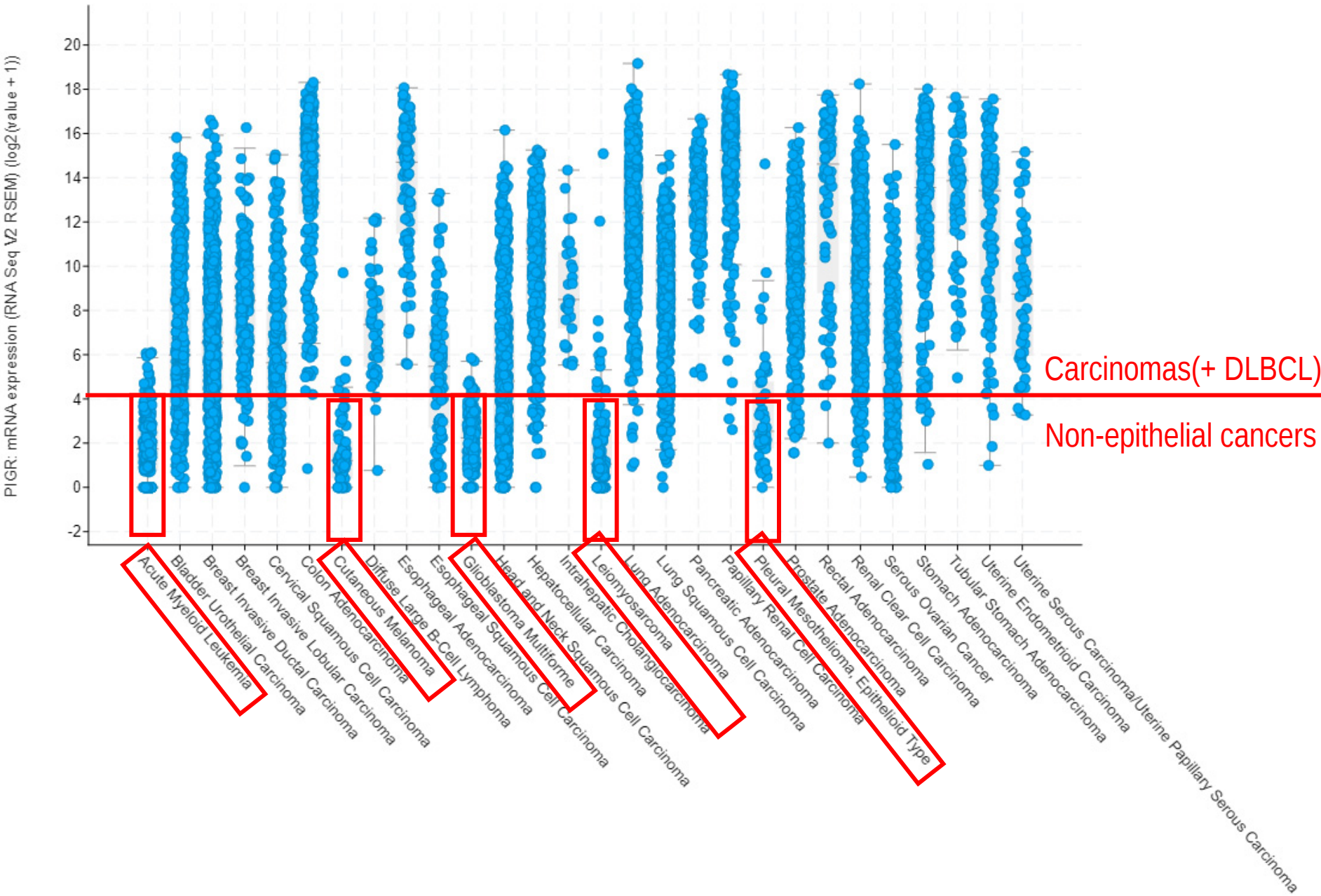
Why irrelevant IgA has anti-tumor activity in an Fc-dependent manner?

pIgR binds polymeric IgA for transcytosis through epithelial cells and luminal secretion

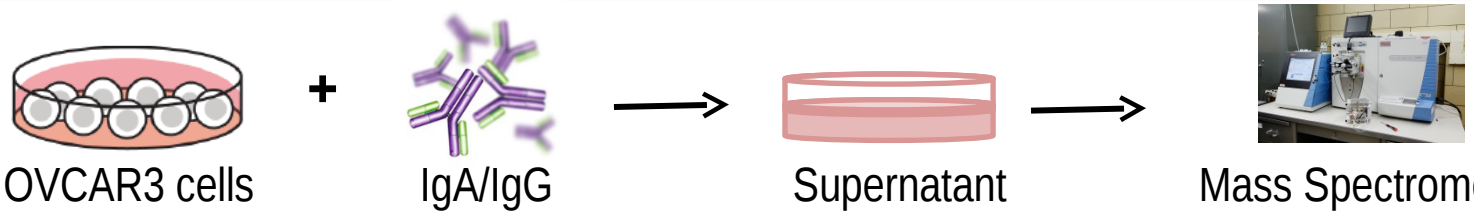


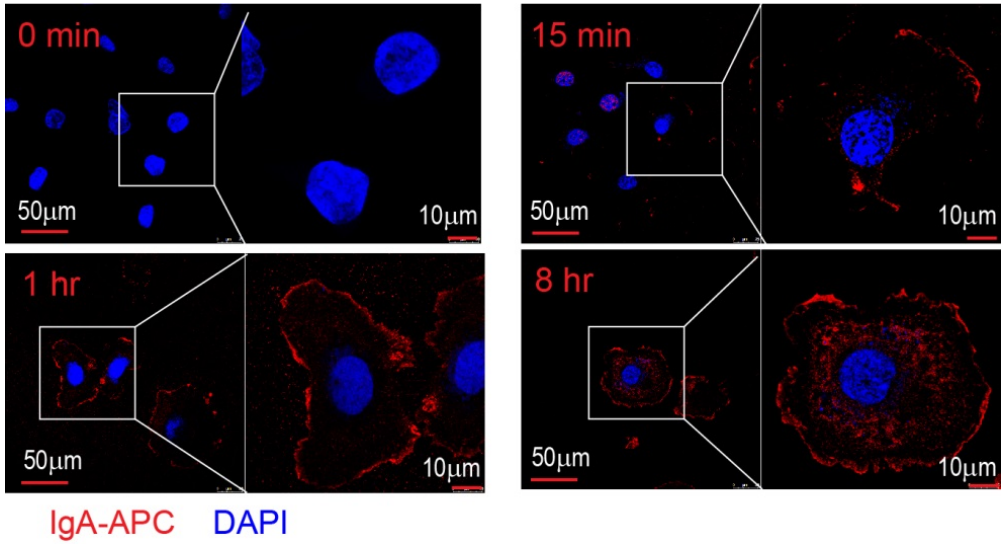


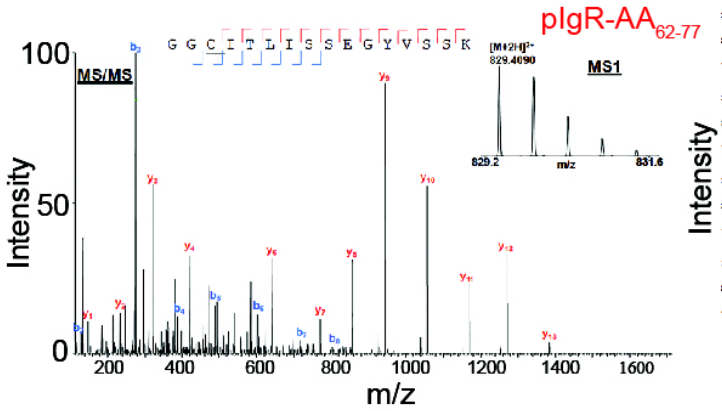
Most epithelial cancers express PIGR (TCGA)

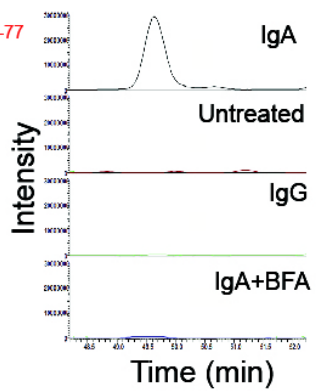


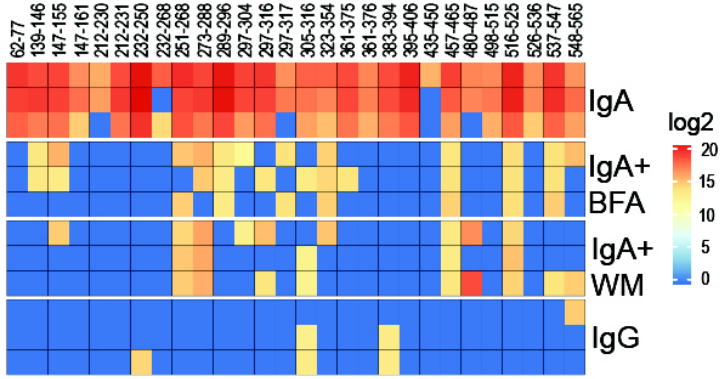
IgA transcytoses inside pIgR+ ovarian cancer cells (release of secretory component upon incubation with IgA)





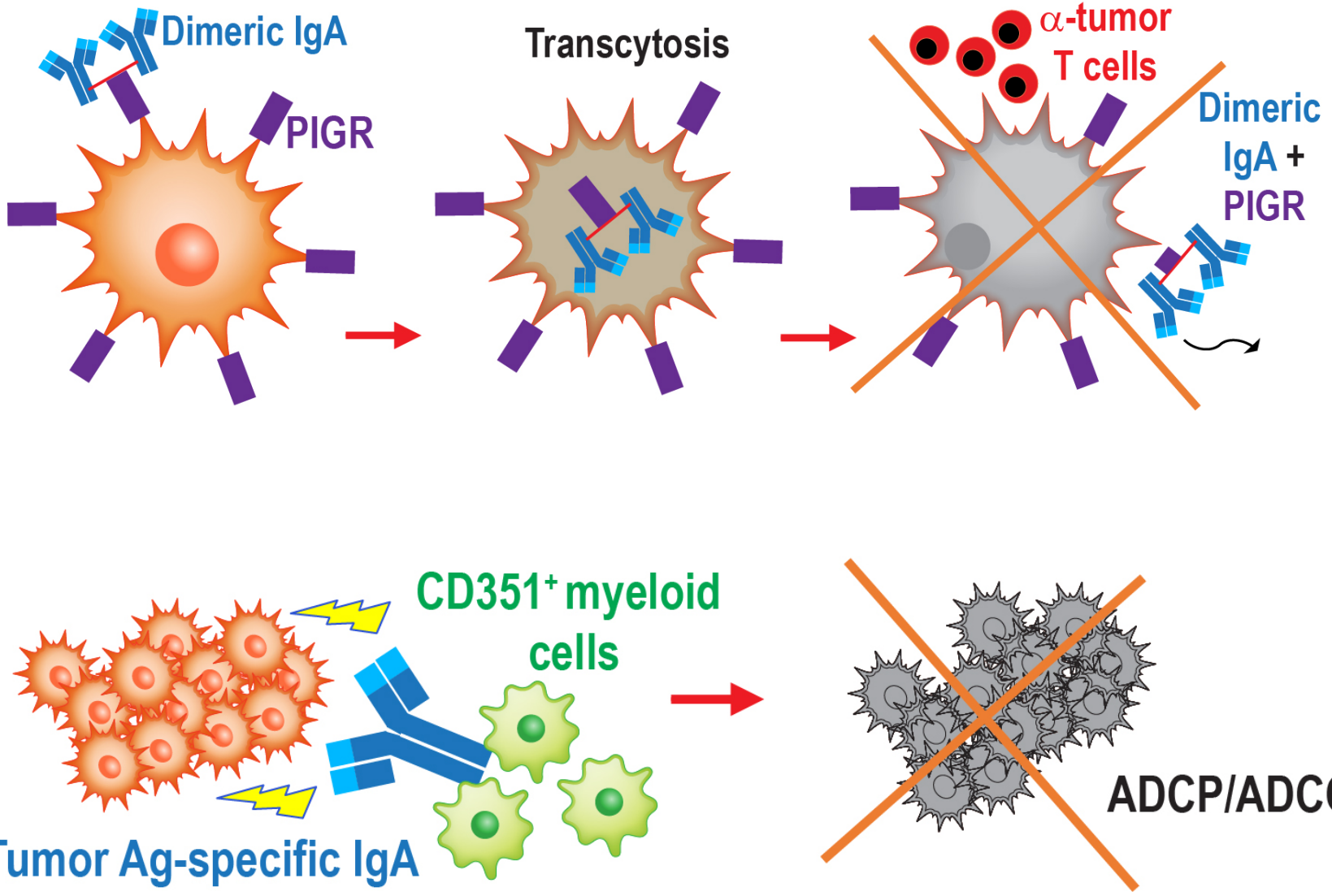






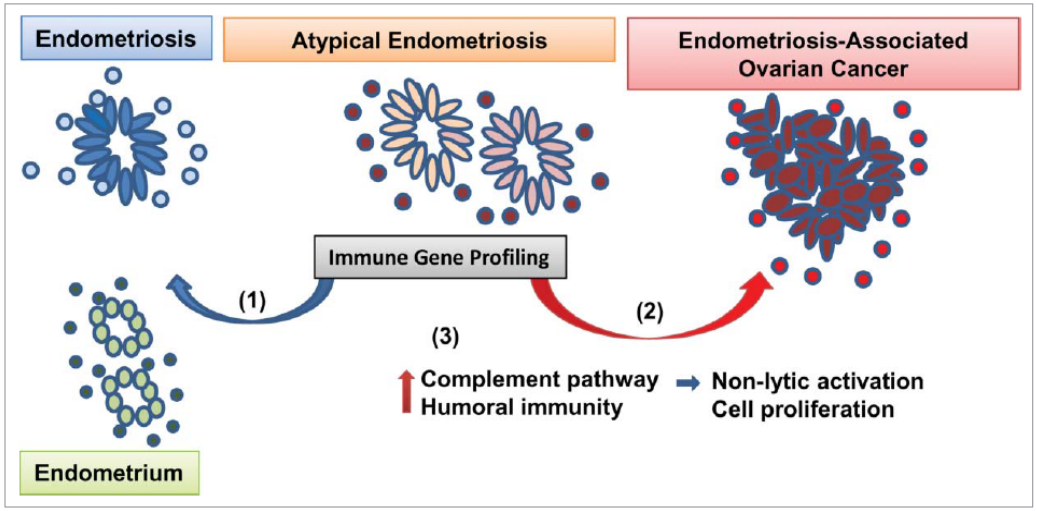
Biswas et al; Nature; 2021

Tumor-derived IgA antibodies elicit protective immune responses through antigen-independent and antigen-dependent mechanisms



What about other histologocal subtypes of ovarian cancer?

Endometriosis is a premalignant lesion for aggressive ovarian cancers



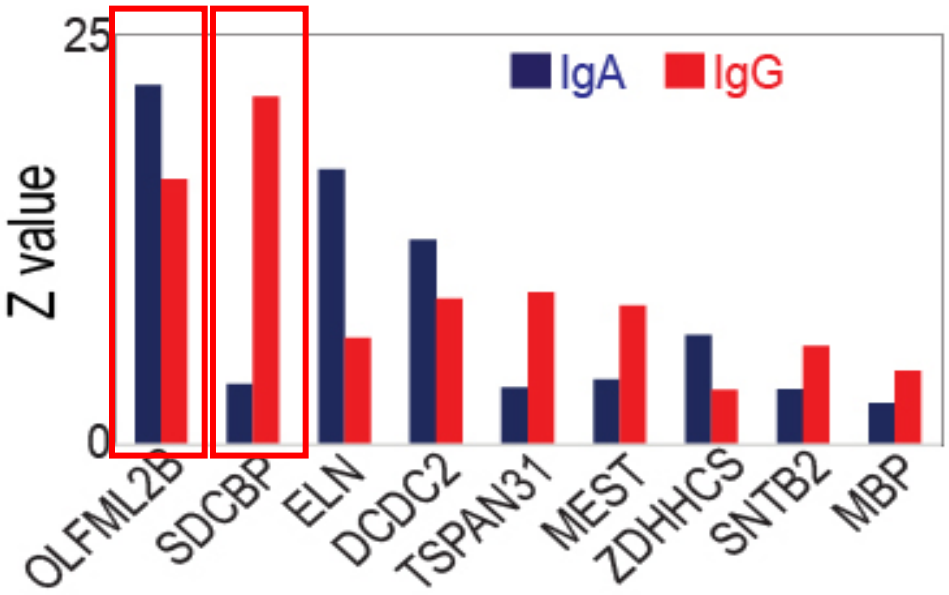
-1/10 women will develop endometriosis during their reproductive years

-10-fold increase in probability of developing clear cell ovarian cancer (5-10% of EOC in Western world; 25% in Asia)

-5-fold increase in probability of developing endometrioid ovarian cancer (8-15% of EOC)

-Increased probability of developing HGSOC (~70% of EOC)

Molecules with extracellular domains targeted by Abs derived from endometriosis and endometriosis-associated ovarian cancers



OLFML2B

-Amplified in ~5% of other gyn/onc malignancies

-Reported association with short survival in gastric cancer

-Co-expressed with ADAM family metallopeptidases, fibrillar collagen proteins and the fibroblast protein FAP→orchestration of stroma and the extracellular matrix.

SDCBP

-Elevated in a wide range of cancers, including melanoma and glioblastoma.

-Links syndecan-mediated signaling to the cytoskeleton→Metastatic driver.

-Drives autophagy to prevent anoikis.

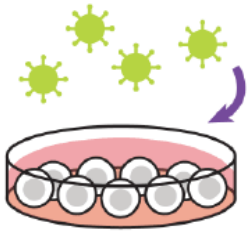
-Promotes chemoresistance in colo-rectal cancer.

2 endometriomas (Ponce, PR), 2 stage III clear cell ovarian cancers, 2 stage III endometrioid ovarian cancers

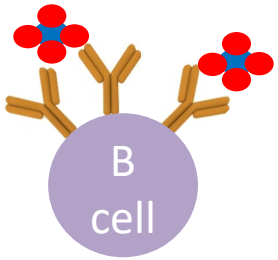
Generating CAR T cells using scFv sequences from tumor-derived Abs

Generation of an OR5V1 CAR from BCR sequences clonally expanded at tumor beds

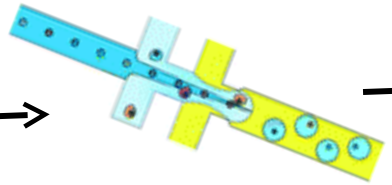
Tumor-derived immortalized B cells



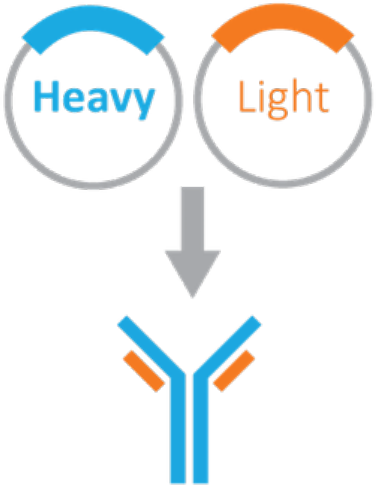
OR5V1-reactive B cell sorting



Droplet encapsulation



scBCR-seq



Chimeric antigen receptor against OR5V1:
Signal peptide; VH; Spacer; VL; CD8 hinge domain; CD8 TM domain; 4-1BB IC domain; CD3zeta

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                                                M  D  W  T  W  S  I  L  F  L  V  A  A  A  T  G  A  H  S  Q
    1 ATGGACTGGACCTGGAGCATCCTTTTCTTGGTGGCAGCAGCAACAGGTGCCCACTCCCAG
                                                V  Q  L  L  Q  S  G  A  E  V  K  K  P  G  A  S  V  K  V  S
   61 GTTCAGCTGCTGCAGTCTGGAGCTGAGGTGAAGAAGCCTGGGGCCTCAGTGAAGGTCTCC
                                                C  K  A  S  G  Y  T  F  T  S  Y  G  I  T  W  V  R  Q  A  P
  121 TGCAAGGCTTCTGGTTACACCTTTACCAGCTATGGTATCACCTGGGTGCGACAGGCCCCT
                                                G  Q  G  L  E  W  M  G  W  I  S  A  Y  N  G  N  T  N  Y  A
  181 GGACAAGGGCTTGAGTGGATGGGATGGATCAGCGCTTACAATGGTAACACAAACTATGCA
                                                Q  K  L  Q  G  R  V  T  M  T  T  D  T  S  T  S  T  A  Y  M
  241 CAGAAGCTCCAGGGCAGAGTCACCATGACCACAGACACATCCACGAGCACAGCCTACATG
                                                D  L  R  S  L  R  S  D  D  T  A  V  Y  Y  C  A  R  A  S  V
  301 GACCTGAGGAGCCTGAGATCTGACGACACGGCCGTGTATTACTGTGCGAGAGCATCCGTG
                                                P  A  P  I  T  G  T  I  I  W  F  D  P  W  G  Q  G  T  L  V
  361 CCGGCCCCTATAACTGGAACTATCATCTGGTTCGACCCCTGGGGCCAGGGGACGCTGGTC
                                                T  V  S  S  G  G  G  G  S  G  G  G  G  S  G  G  G  G  S  G
  421 ACCGTCTCCTCAGGTGGAGGCGGTTCAGGCGGCGGTGGCTCTGGCGGTGGCGGATCGGGC
                                                S  V  T  S  Y  V  L  T  Q  P  P  S  V  S  V  A  P  G  Q  T
  481 TCTGTGACCTCCTACGTGCTGACTCAGCCACCCTCGGTGTCAGTGGCCCCAGGACAGACG
                                                A  R  I  T  C  G  G  N  N  I  G  S  K  S  V  H  W  Y  Q  Q
  541 GCCAGGATTACCTGTGGGGGAAACAACATTGGAAGTAAAAGTGTACACTGGTACCAGCAG
                                                K  P  G  Q  A  P  V  L  V  V  Y  D  D  S  D  R  P  S  G  I
  601 AAGCCAGGCCAGGCCCCTGTGCTGGTCGTCTATGATGATAGCGACCGGCCCTCAGGGATC
                                                P  E  R  F  S  G  S  N  S  G  N  T  A  T  L  T  I  S  R  V
  661 CCTGAGCGATTCTCTGGCTCCAACTCTGGGAACACGGCCACCCTGACCATCAGCAGGGTC
                                                E  V  V  F  G  G  G  T  K  L  T  V  L  K  V  E  N  H  T  A
  721 GAAGTGGTATTCGGCGGAGGGACCAAGCTGACCGTCCTAAAGGTGGAAAACCACACCGCC
                                                C  H  C  S  T  C  Y  Y  H  K  S  A  S  T  T  T  P  A  P  R
  781 TGCCACTGCAGCACATGCTACTACCACAAGAGCGCTAGCACCACCACCCCTGCCCCTAGA
                                                P  P  T  P  A  P  T  I  A  S  Q  P  L  S  L  R  P  E  A  C
  841 CCTCCAACACCCGCCCCTACAATCGCCTCCCAGCCTCTGTCTCTGAGGCCCGAGGCTTGT
                                                R  P  A  A  G  G  A  V  H  T  R  G  L  D  F  A  C  D  I  Y
  901 AGACCAGCTGCTGGCGGAGCCGTGCACACCAGAGGACTGGATTTCGCCTGCGACATCTAC
                                                I  W  A  P  L  A  G  T  C  G  V  L  L  L  S  L  V  I  T  L
  961 ATCTGGGCCCCTCTGGCCGGCACATGTGGCGTGCTGCTGCTGAGCCTCGTGATCACCCTG
                                                M  C  K  R  G  R  K  K  L  L  Y  I  F  K  Q  P  F  M  R  P
 1021 TACTGCAAGCGGGGCAGAAAGAAGCTGCTGTACATCTTCAAGCAGCCCTTCATGCGGCCC
                                                V  Q  T  T  Q  E  E  D  G  C  S  C  R  F  P  E  E  E  E  G
 1081 GTGCAGACCACCCAGGAAGAGGACGGCTGCTCCTGCAGATTCCCCGAAGAGGAAGAGGGG
                                                G  C  E  L  R  V  K  F  S  R  S  A  D  A  P  A  Y  K  Q  G
 1141 GGCTGCGAACTGAGAGTGAAGTTCAGCAGAAGCGCCGACGCCCCTGCCTACAAGCAGGGC
                                                Q  N  Q  L  Y  N  E  L  N  L  G  R  R  E  E  Y  D  V  L  D
 1201 CAGAACCAGCTGTACAACGAGCTGAACCTGGGCAGACGGGAAGAGTACGACGTGCTGGAC
                                                K  R  R  G  R  D  P  E  M  G  G  K  P  R  R  K  N  P  Q  E
 1261 AAGCGGAGAGGCAGGGACCCTGAGATGGGCGGAAAGCCCAGACGGAAGAACCCCCAGGAA
                                                G  L  Y  N  E  L  Q  K  D  K  M  A  E  A  Y  S  E  I  G  M
 1321 GGCCTGTATAACGAACTGCAGAAAGACAAGATGGCCGAGGCCTACAGCGAGATCGGAATG
                                                K  G  E  R  R  R  G  K  G  H  D  G  L  Y  Q  G  L  S  T  A
 1381 AAGGGCGAGCGGAGAAGAGGCAAGGGCCACGATGGCCTGTACCAGGGCCTGAGCACCGCC
                                                T  K  D  T  Y  D  A  L  H  M  Q  A  L  P  P  R  *
 1441 ACCAAGGACACCTATGACGCCCTGCACATGCAGGCCCTGCCCCCCAGATAA 1491
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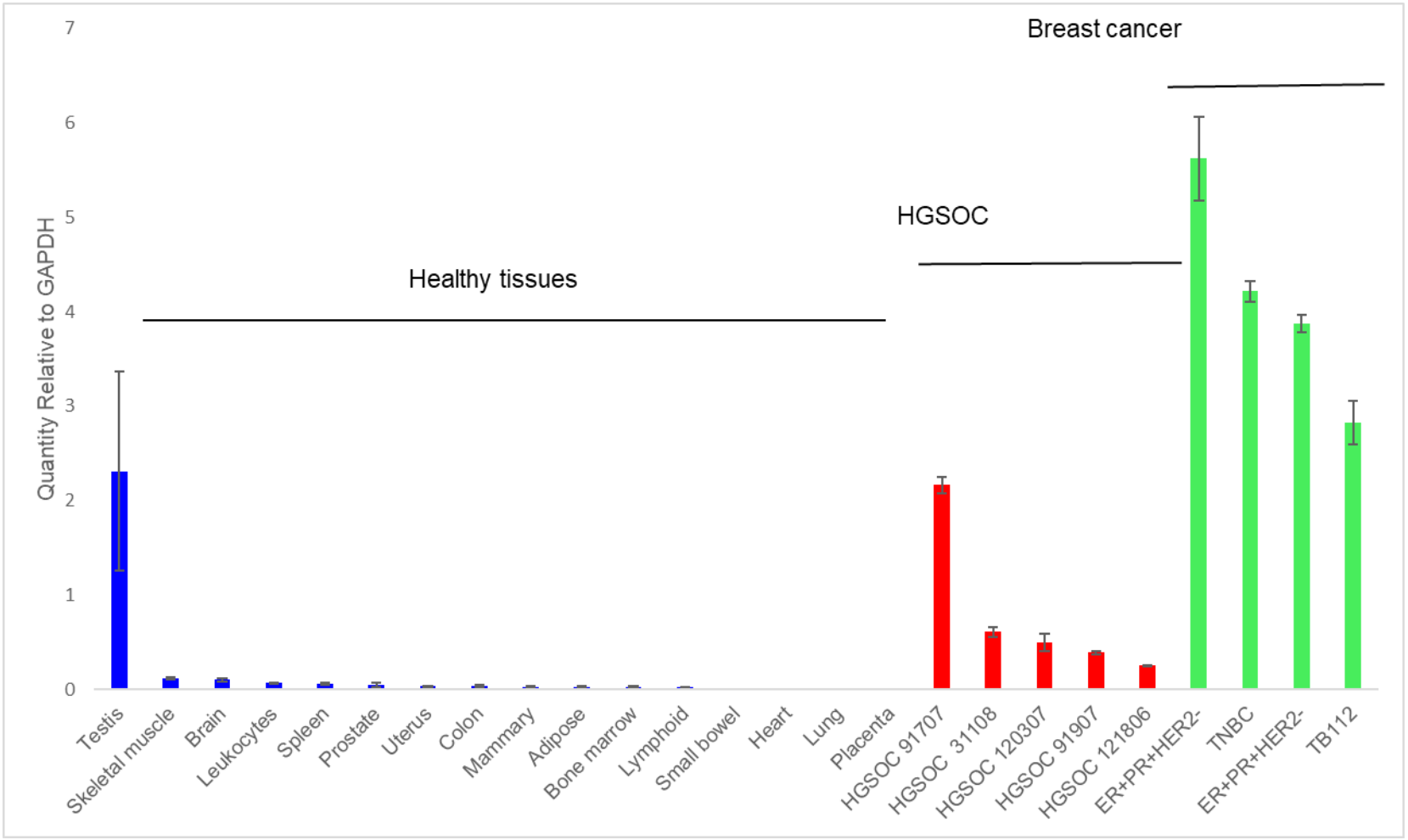
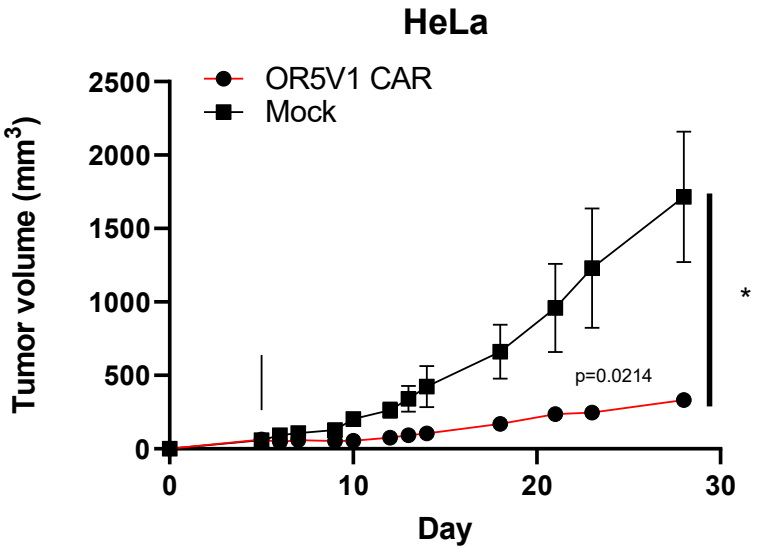
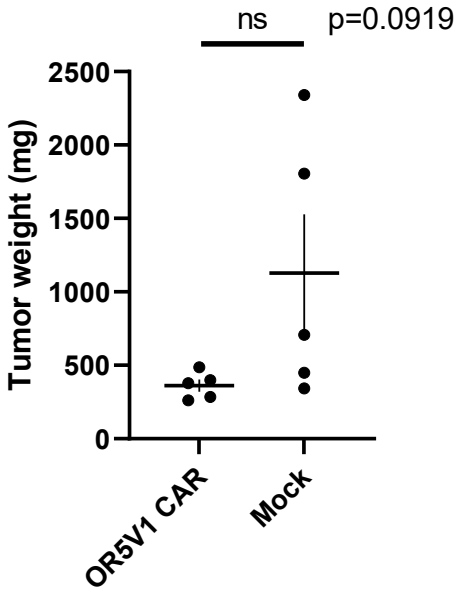
Targeting OR5V1 with CAR T cells that use a scFV derived from ovarian cancer Abs

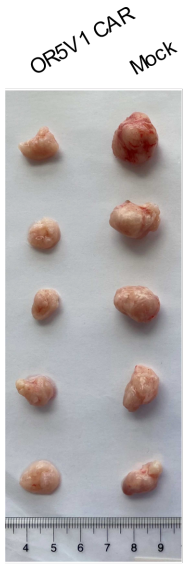


Tumor-derived OR5V1 CAR T cells abrogate HeLa tumor growth in vivo

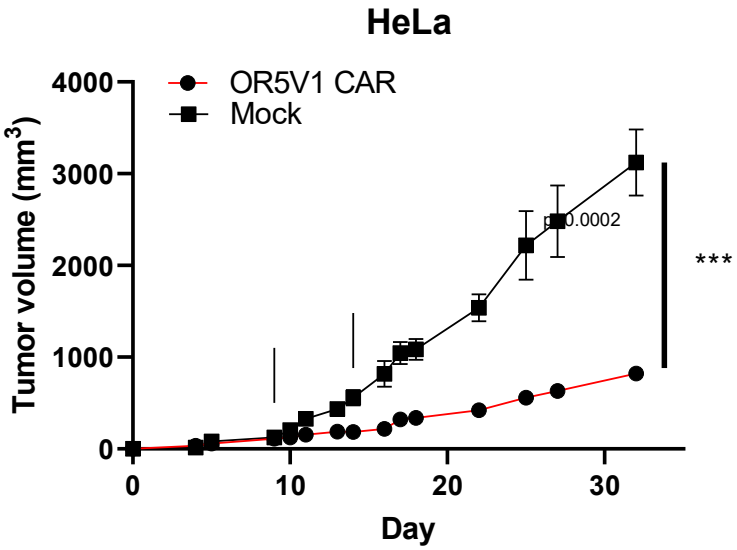
Day 5 injection

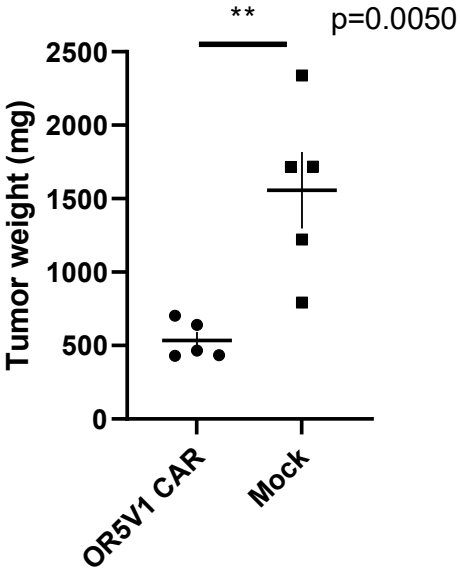


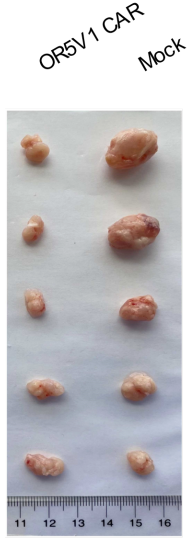




Days 9&14 injections





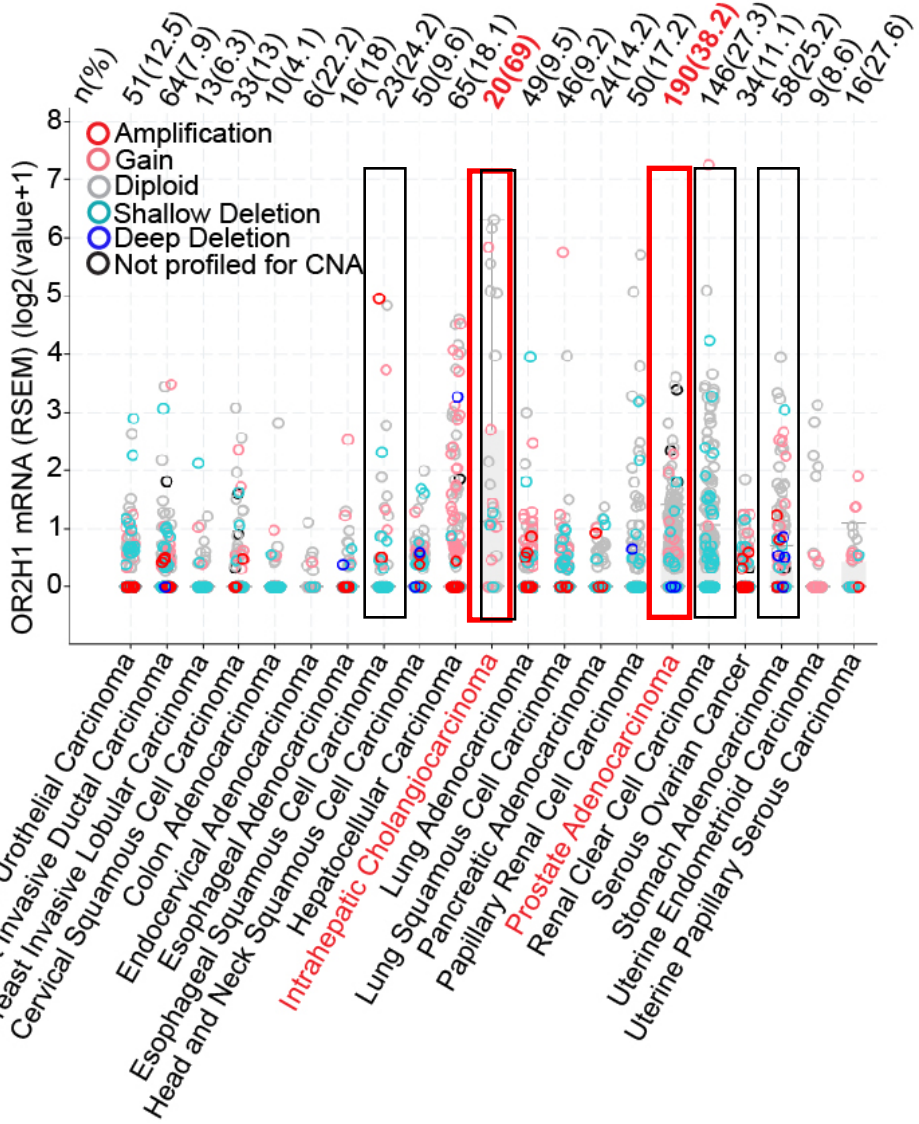


In vivo experiment 2, n=5 NSG mice/group

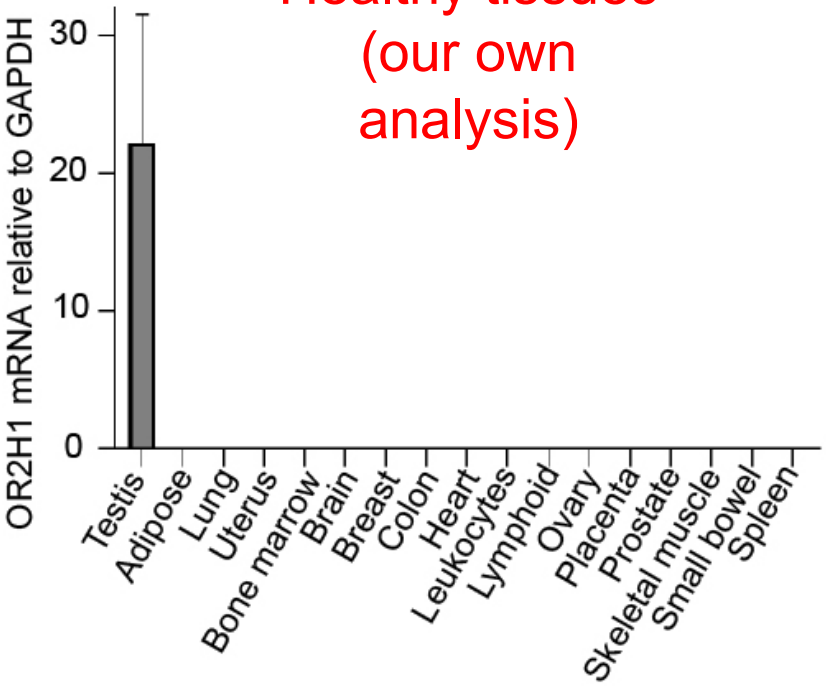
Generating CAR T cells against other olfactory receptors (OR2H1)

OR2H1 is expressed in multiple human tumors, but not most healthy tissues (with the exception of testis)

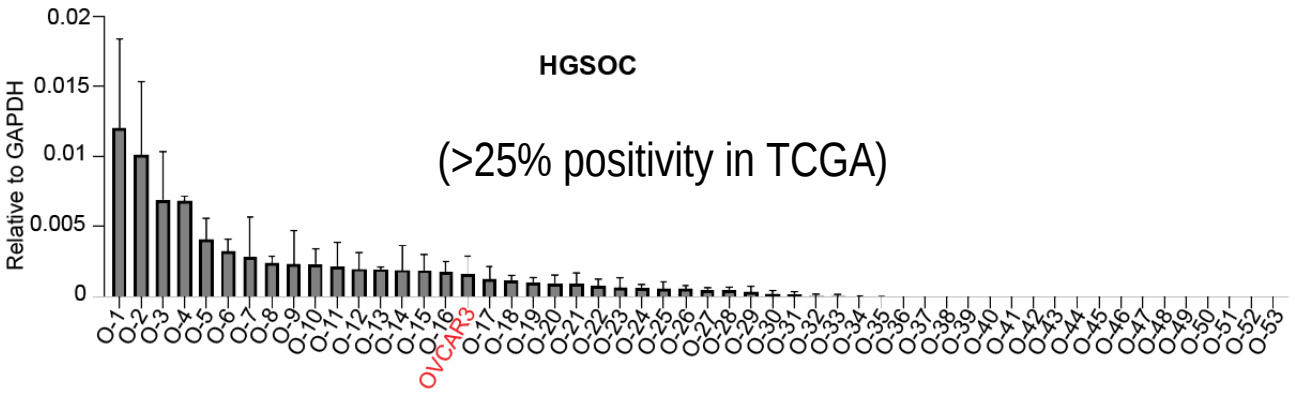
TCGA (7%-69% of carcinomas)

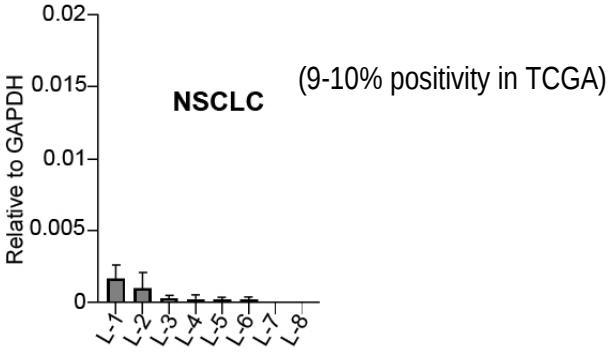


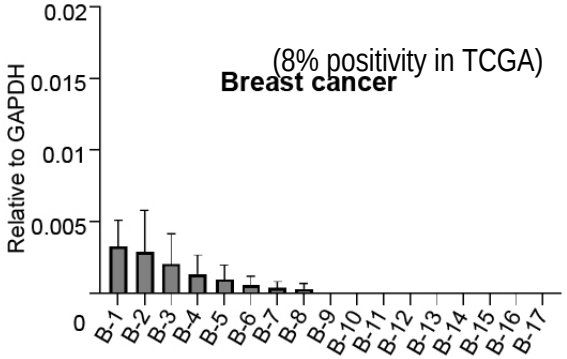
Healthy tissues (our own analysis)

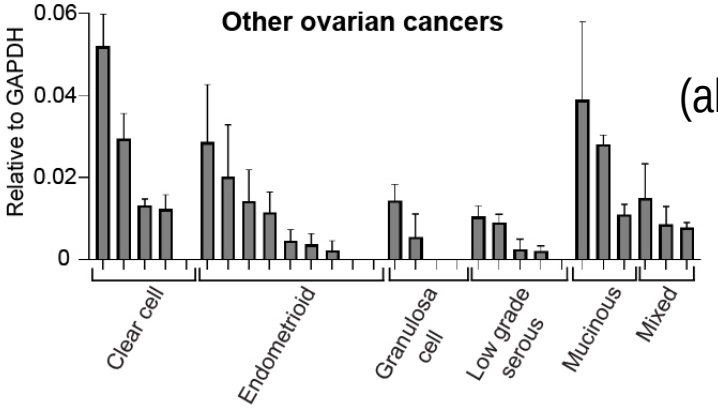


OR2H1 is expressed in human ovarian cancer

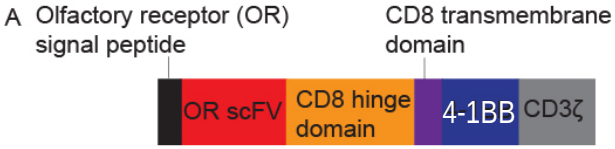


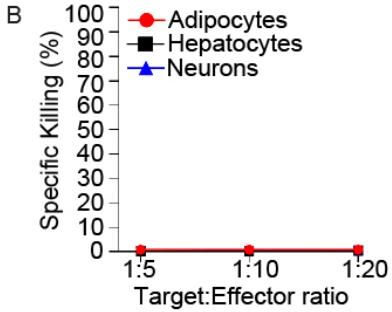




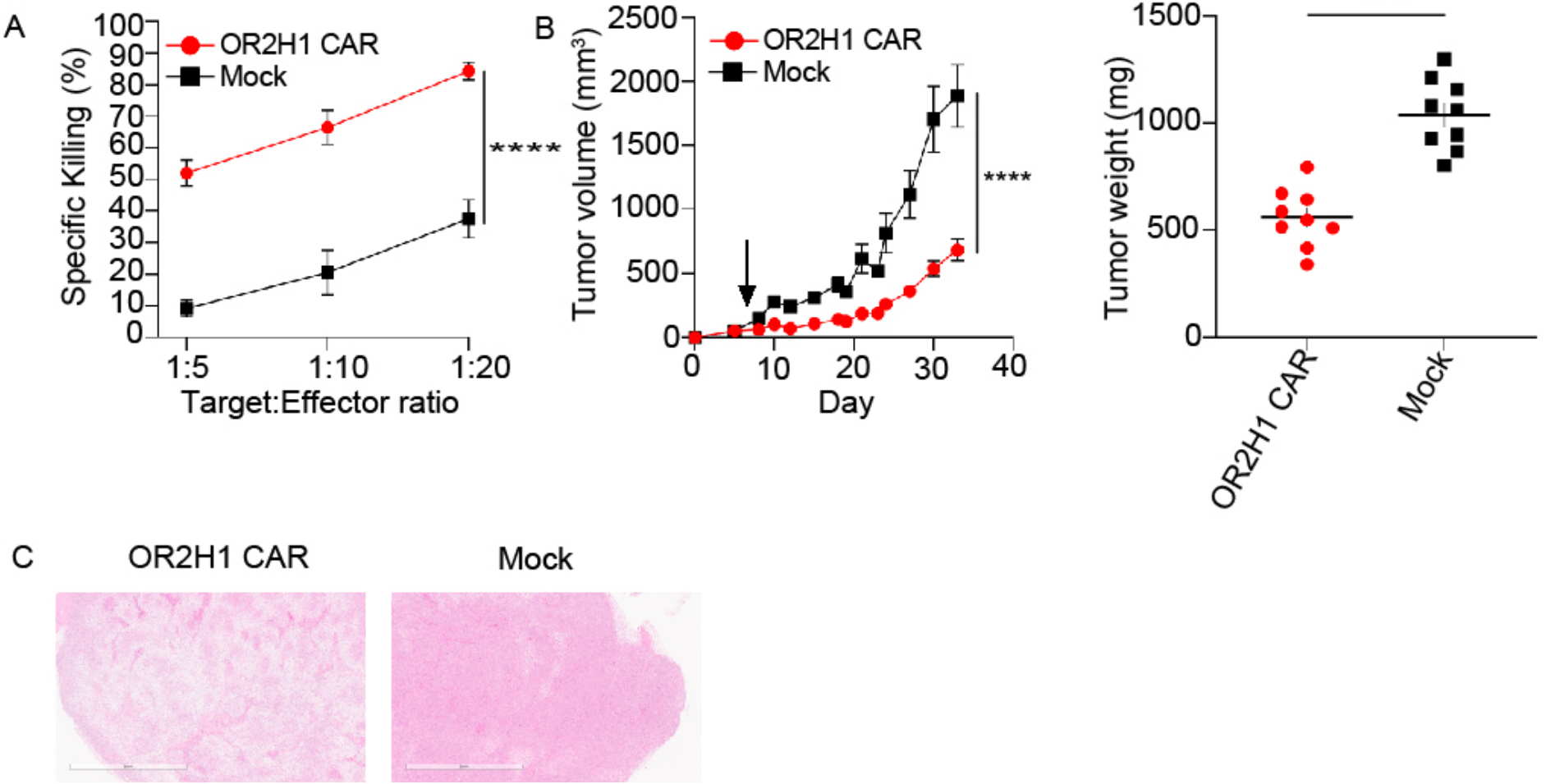


OR2H1 CAR T cells abrogate NSCLC growth in vivo



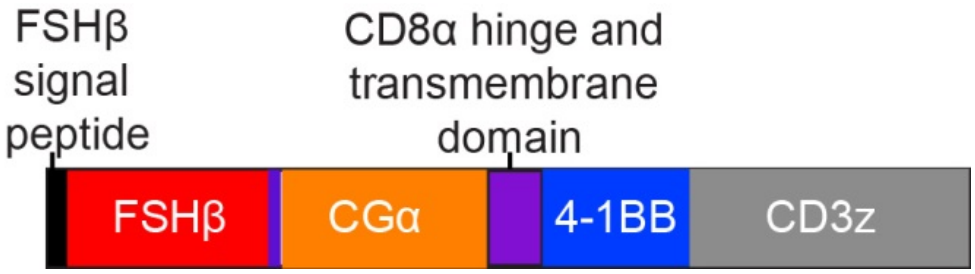


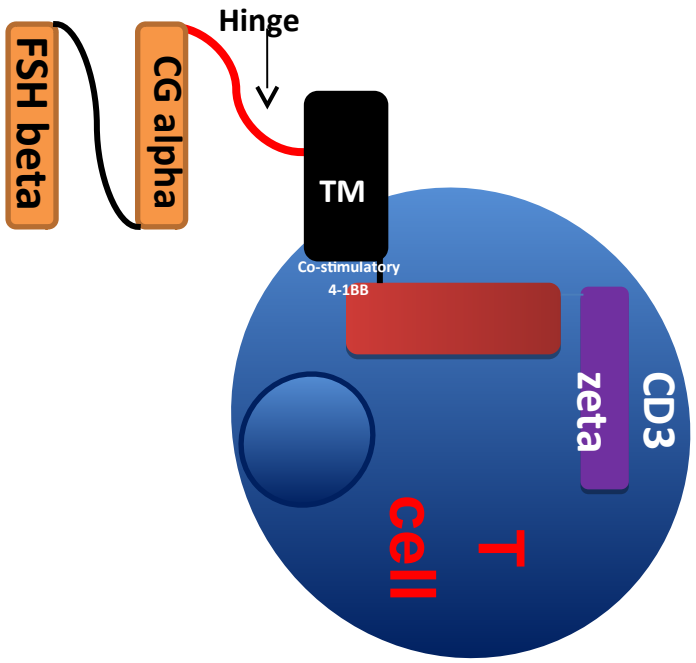
OR2H1 CAR T cells control OR2H1^{low} OVCAR3 (HGSOC) tumor growth



IND approval for FSHCER T cells targeting FSHR+ ovarian cancer (IND:27225)

FSH-targeted chimeric receptors re-direct primary human T cells against FSHR+ ovarian cancer cells



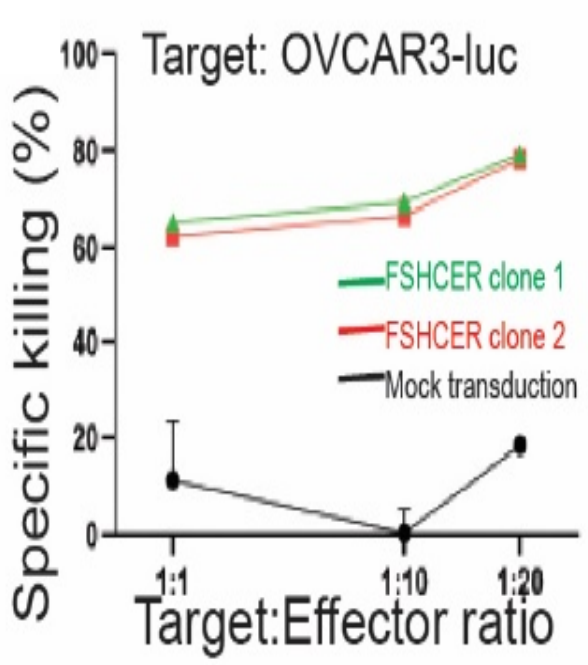


IND# 27225:

Autologous CD3+T cells transduced w/ gamma retroviral vector, pMSGV1, for FSHR-specific 4-1BB/CD3 chimeric endocrine receptor expression, Intraperitoneal (IP)/Intravenous (IV) infusion

FSH-re-directed autologous human T cells effectively target orthotopic patient-derived FSHR+ tumors in vivo (II)

10e7 AUTOLOGOUS CERT cells IP



IND# 27225:

Autologous CD3+T cells transduced w/ gamma retroviral vector, pMSGV1, for FSHR-specific 4-1BB/CD3 chimeric endocrine receptor expression, Intraperitoneal (IP)/Intravenous (IV) infusion

CONCLUSIONS

→T and B cell responses act in coordination in HGSOC.

→IgA elicits antigen-specific and non-antigen-specific (PIGR-dependent) anti-tumor effects in EOC.

→Tumor-derived Abs targeting extracellular molecules abrogate ovarian cancer progression.

→CAR T cells against ovarian cancer can be generated from BCR sequences clonally expanded at tumor beds.

→OR2H1 CAR T cells can target a variety of human cancers.

→FSH-targeted T cells expressing chimeric receptors kill established ovarian tumors

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U01CA232758 (NCI)
DoD Ovarian Cancer Academy

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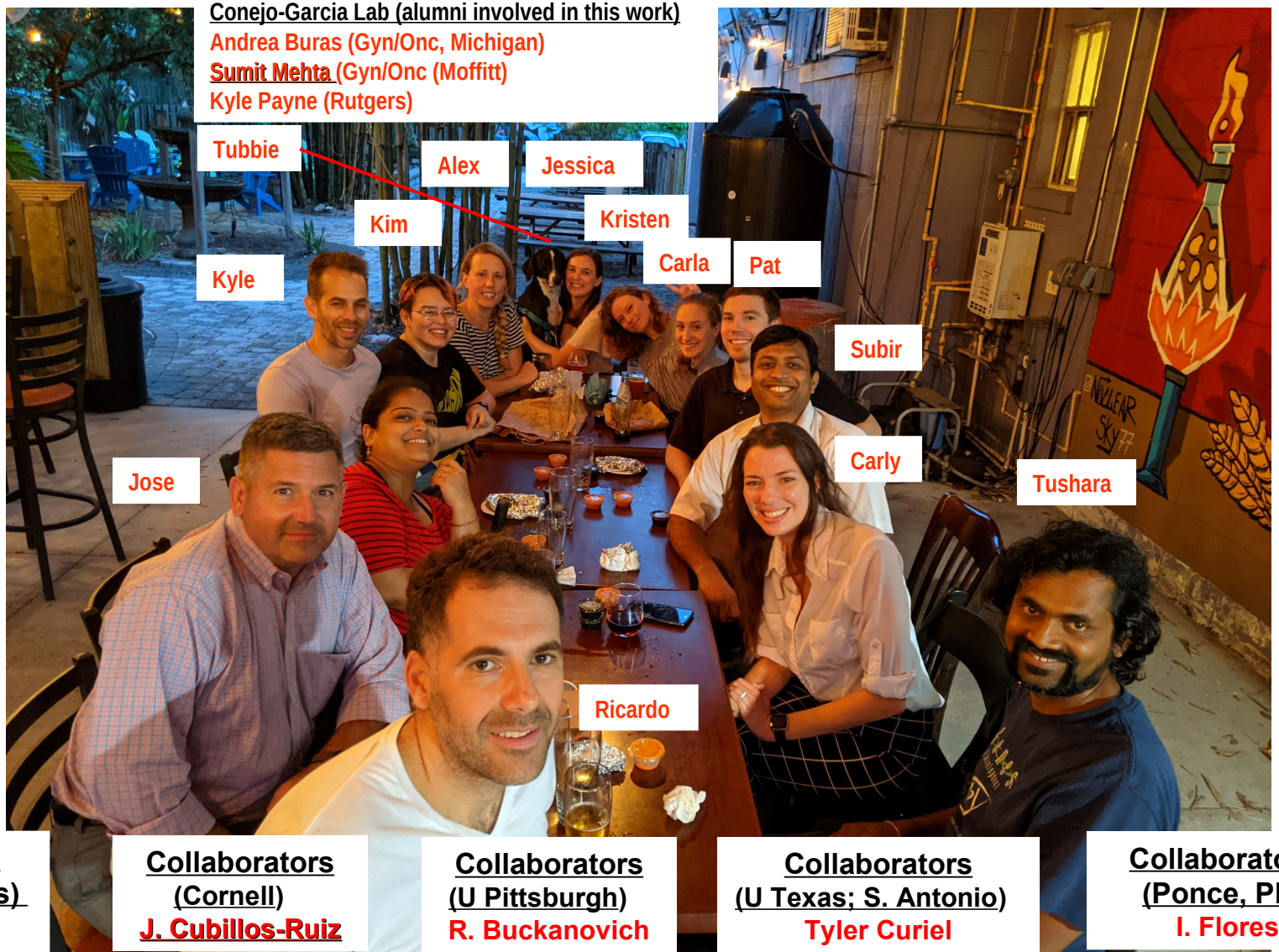

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